



Article

Exploration of the Benefits of Biofertilizers for Attaining Food Security in Egypt's Agriculture

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Abstract: Biofertilizers and biocontrol agents have been improved for several decades as microbiological tools that can provide beneficial outcomes in the growth and health of plants. Two field experiments were performed in the Scientific Farm of the Horticulture Department, Faculty of Agriculture, Minia University, Egypt during the winter of 2022/2023 using clay loam soil. Control plots were treated with the recommended rates of mineral fertilizer of NPK (100%) without inoculation, while other plots were inoculated with Egyptian isolates of effective microorganisms (EMs) (a mixture of *Azotobacter chroococcum* and *Azospirillum brasilense*) and strains of *Bacillus* spp.; the biofertilizers were applied to the soil through mixing with irrigation water. For mycorrhizae, *Glomus mosseae* and *Glomus fasciculatum* isolation stock cultures were combined to create the mycorrhizal inoculum. The results showed that biofertilizers with 75% NPK were the best. Biofertilizers changed the properties of soil, increased its content of beneficial microorganisms, increased the total good quality production of onion and potato and decreased the stress of chemical pesticides and mineral fertilizers on crop growth and productivity.

Keywords: mycorrhizae; biofertilizers; EM; potato; onion; crop production



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1. Introduction

The global population will reach about 9 billion by 2050 according to the Food and Agricultural Organization; therefore, there must be an enhancement in crop production to meet the increasing food demand [1]. Food security problems will be increased by climate change in many countries [2]. Many of the direct negative effects on food security are attributed to modifications in soil characteristics, water availability and crop production. Soil represents a substantial source of food productivity for many humans throughout their lifespan. Direct negative effects on the quality of soil can be caused by the fertility depletion of soil, the reduction in soil organic matter, the decrease in available soil water-holding capacity and the degradation in soil structure, erosion, salinization and crusting. There are also many indirect effects of climate change, e.g., variations in cropping/farming systems [3].

Onion and potato were selected in this research, as both are important crops in Egypt and all over the world. Potato (*Solanum tuberosum*) is a perennial plant which belongs to

the non-grain food group [4]. Globally, potato is a common tuber crop and the fourth most important crop after the master cereal crops rice, wheat and maize [5,6]. It is considered a main source of dietary fibers, complex carbohydrates, proteins and vitamins and also contains essential minerals such as manganese calcium, copper, potassium, iron and zinc [7]. More than half of the overall potato production takes place in developing countries such as Egypt [8]. Every year, about 388,191 thousand tons of potato are produced worldwide, ensuring food supply stability [9]. Onion (*Allium cepa* L.) is one of the most substantial commercial crops, used as a vegetable, spice and in condiments throughout the world including Egypt [10]. It is widely used throughout the year for cooking with other types of vegetables, in addition to its benefits in many medical uses [11]. It is highly valued as a plant material that can be stored for eight to ten months and as a flavoring agent [12]. In Egypt, the area cultivated with onion was estimated as 82760 hectares [13]. Middle Egypt produces millions of tons of onion (green, cured and processed) and potato (fresh and processed) every year, and most of these products are exported to many countries in Africa, Europe, the US and the gulf countries.

In the past decades, due to the rise in agricultural practices such as pesticides and chemical fertilizers, agriculture has been degraded at a global scale, and breeds possess lower fertility due to the lack of biodiversity, disturbances in biogeochemical cycles and water retention issues. Both plant productivity and soil health are roughly influenced by many interactions among soil, plants and microorganisms [14]. Soil microbes collaborate with one another and with plant roots to provide a wide variety of substantial acts that are valuable for sustaining the ecological balance of the soil [15]. The interactions of plant microbes are considered positive if they can improve plant survival, crop productivity and nutritional status, and they are considered negative if they decrease plant growth. Soil fertility is inextricably related to the balance of plants and microorganisms [16].

Microbiological tools, which include biocontrol and biofertilizer agents, as well as fungi and bacteria, which can provide valuable outcomes in both the growth and health of plants, have been improved for many decades. Biofertilizers have the power to ameliorate crop productivity through many environmentally friendly mechanisms [17]. On the other hand, biofertilizers contain living microorganisms that induce plant growth through colonizing the rhizosphere or the internal tissues of the plant when applied to surfaces of plants, soil or seeds. Biofertilizers are capable of phosphate solubilization, nitrogen fixation, sulfur oxidization, decomposition of organic compounds or plant hormone production [18]. Generally, biofertilizers perform nutrient cycling and assure optimal development and growth of crops [19]. On the other hand, biocontrol agents are potentially able to replace the harmful effect of pesticides. Biological control also supplies a non-chemical method for controlling plant diseases through using other living entities, e.g., microorganisms. The ability of microbes as a biocontrol can be a result of producing antibiotic enzymes or compounds that can cause iron depletion from the rhizosphere, lysis of the fungal cell wall and induction of systemic resistance [20]. The application of biofertilizers is a possible approach to enhancing soil microbial status that motivates the natural soil microbiota, thus affecting nutrient accessibility and organic matter decomposition [21]. The ability of biofertilizers to create a high level of microbial diversity in soil could result in better crop output for sustainable agriculture [22].

Biological control agents can also be used to treat plant diseases as alternatives to synthetic pesticides because of their perceived enhanced stage of safety, lower environmental implications and capability to reduce disease whilst being less harmful than conventional fungicides [23]. Recently, it has been confirmed that biological control agents are a useful technique for fighting plant infections [24].

This paper provides information on efficient approaches such as biofertilizers which can improve the restoration of agricultural soil, therefore enhancing crop health to attain sustainable agriculture. We focused on producing some safe and clean crop products through biocontrol of some soilborne diseases of some crops, as there are few studies on plant disease biocontrol in Egypt. Moreover, the research aims to study the impacts of

these biofertilizers, biocontrol microorganisms and mineral fertilizers (NPK) on soil and microbiological properties of the soil. This can enable agriculturalists to improve farming and reach high standards of soil quality, which subsequently leads to improved plant development.

2. Materials and Methods

2.1. Studied Plant Species

Two field experiments were established in the Scientific Farm of the Horticulture Department, Faculty of Agriculture at Minia University, Egypt (28.11 N, 30.11 E) (Figure 1) during winter 2022/2023. The study was conducted to investigate the effects of using various biocontrol mycorrhizae biofertilizer (EMs) on the growth and production of safe and economic onion yield of bulbs (Italian Red cv.) and potato yield of tubers (Cara cv.) along with decreasing the negative effects of climate change on the growth and production of these two important crops. Onion and potato were selected as both are important crops in Egypt and worldwide.

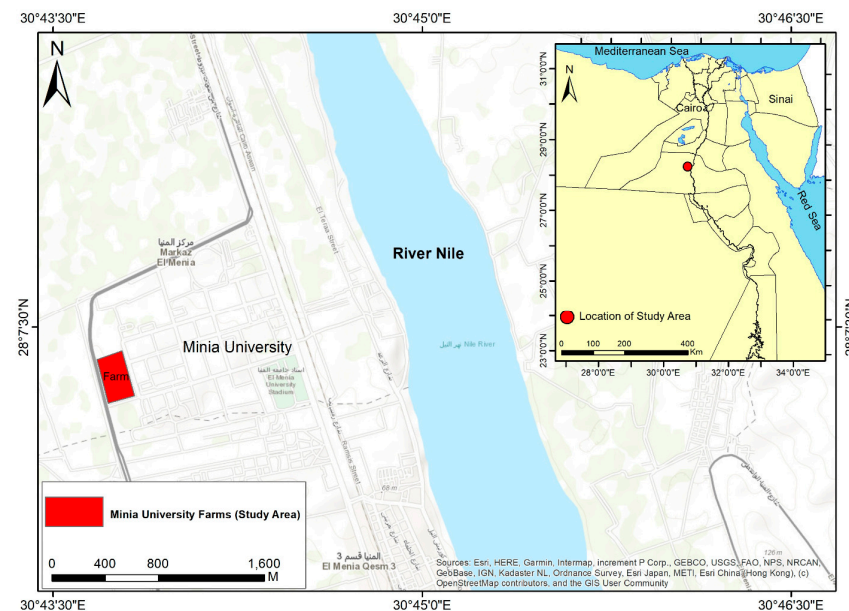


Figure 1. Map of Minia city showing the location of Minia University Farm (place of study), Egypt.

Potato tubers (Cara cv.) used in this study were planted on 15 October 2022, 20 cm apart and on one side of ridges. Throughout the experiment, soil moisture was kept at field capacity by watering every 8–12 days through a furrow irrigation system. On the other hand, onion seedlings (cv. Italian Red) were planted on 1 October 2022 on the top of ridges with one line/ridge and with 15 cm between the seedlings. The seedlings were planted in dry soil, and then the plots were irrigated with a furrow irrigation system. All other good agricultural practices (GAPs) were carried out as recommended by the Egyptian Ministry of Agriculture and Soil Reclamation (EMASL) for potato and onion crop production.

2.2. Soil and Biofertilizers

The soil used in the experiment was clay loam, collected from the surface layer of the experimental field. The physical and chemical properties of the soil were analyzed before the experiment (Table 1). Egyptian isolates of effective microorganisms (EMs) nitrogen-fixing bacteria products were used as a mixture of *Azotobacter chroococcum* and *Azospirillum brasilense*. Phosphate-dissolving bacteria and potassium-releasing bacteria were used, specifically the strains of *Bacillus megaterium* and *Bacillus circulans*, respectively. All biofertilizers were supplied by the department of Agricultural Microbiology, Faculty of Agriculture, Minia University, Egypt. The biofertilizers were applied to the soil through

mixing with irrigation water at the rate of 11.9 L ha^{-1} in two equal doses after 30 and 60 days from the planting date (10 October 2022) by using liquid cultures (1 mL contains 10^8 cell). For mycorrhizae, *Glomus mosseae* and *Glamus fasciculatum* isolation stock cultures were combined to create the mycorrhizal inoculum. The process of mycorrhizal inoculation involved mixing 100 g of a mycorrhizal inoculum from our stock culture collection to each pot. These well-mixed rhizosphere samples were kept in polyethylene bags at 4°C for 3–6 months before being added to the soil. The samples included spores, hyphae and mycorrhizal root fragments (80% root colonization). Plots (control) of soil were left uninoculated so that they could be compared with treatments as a control. Four treatments were taken during the experiment according to Table 2.

Table 1. Physicochemical characteristics of the soil used in the experiments prior to planting with onion and potato crops.

Attribute	Value
Sand (%)	41.62
Silt (%)	28.13
Clay (%)	30.25
Texture class	Clay loam
Bulk density (Mg m^{-3})	1.27
pH 1:2.5 (water)	8.15
ECe (dS m^{-1})	5.04
OC (g kg^{-1})	8.4
TN (g kg^{-1})	1.13
ava. P (g kg^{-1})	22
ava. K (g kg^{-1})	206

Table 2. Treatment details.

Treatment	Dose
T1	Mycorrhizae + 75% NPK
T2	EM + 75% NPK
T3	75% NPK
T4	100% NPK

2.3. Experimental Design

A randomized complete block design (RCBD) experiment was performed during winter using three treatments with three replicates for each crop. The experiment was carried out in plots with area 10.5 m^2 (3 m length and 3.5 m width). Each plot included 5 rows (70 cm width of each ridge).

The recommended dose of NPK (100%) as the control treatment for the potato was $357.0 \text{ N kg ha}^{-1}$ (as ammonium nitrate; 33.5% N), $142.8 \text{ kg ha}^{-1} \text{ P}_2\text{O}_5$ (as calcium super phosphate; 15.5% P_2O_5) and $228.5 \text{ kg ha}^{-1} \text{ K}_2\text{O}$ (as potassium sulphate; 48% K_2O) according to the Egyptian Ministry of Agriculture and Soil Reclamation (EMASL). Full doses of P were added to the soil before sowing, while N and K were added in two equal doses, one after 30 days from planting onion seedlings or potato seed-tubers and the other 20 days later.

Seventy-five percent of the recommended doses of N, P and K fertilizers were applied for potato and onion production according to the Egyptian Ministry of Agriculture and Soil Reclamation (EMASL).

The experiment included the use of two doses of NPK, 75% and 100% (control). Seventy-five percent of the recommended doses of N, P and K fertilizers were applied to all pots of potato and onion production except for the control in accordance with the Egyptian Ministry of Agriculture and Soil Reclamation (EMASL). The four treatments were T1 (mycorrhiza + 75%NPK), T2 (EMs + 75% NPK), T3 (75% NPK only) and Ta (100% NPK).

2.4. Data Recorded

At harvest, the following data were recorded for potato: total yield (t ha^{-1}), fresh weight of a single tuber (g) and dry weight of 100 g of fresh weight (g). Tuber rotting and scabs were also determined visually by estimating their percentage on the surface of tubers after harvest. For onion, plant height (cm), total yield (t ha^{-1}), fresh weight (g), dry weight of 100 gm of fresh weight (g), bulb rotting and sprouting were recorded. Bulb rot was determined visually at harvest, while sprouting was determined by observing elongation of leaf blade in a specific size class of leaf sheaths during storage at 18 °C.

2.5. Determination of the Degree of Mycorrhizal Colonization

Roots of potato and onion were mixed to yield approximately 1 g fresh weight, cleansed with water and stored in 70% alcohol prior to the examination of mycorrhizal colonization. After storage, root samples were cut into approximately 1 cm segments, boiled in 15% KOH for 40 min, stained with aniline blue and fixed in 40% lactic acid [25]. The stained roots were suspended in a few drops of lactic acid on glass slides and examined microscopically. The percentage of infection in 100 pieces were calculated as follows:

$$\text{Root colonization \%} = \frac{\text{no. of VAM positive pieces} \times 100}{\text{no. of pieces scored}}$$

where VAM is the vesicular arbuscular mycorrhizae (VAM).

2.6. Total Counts of Bacteria, Fungi and Actinomycetes

Total counts of bacteria fungi and actinomycetes were determined using the plate count technique according to [26]. Colony forming units (CFU) of total bacteria and actinomycetes were counted on nutrient agar medium and the total fungi by using potato dextrose agar medium.

2.7. Soil Analysis

At the end of the experiment, top surface soil samples were collected from each experimental field, dried at 60 °C, ground and sieved to 2 mm then stored in plastic containers at approximately 4 °C. Undisturbed soil samples were collected using stainless-steel rings (5 cm in diameter and 5 cm in height) to determine the bulk density [27]. Particle size distribution was determined using the Bouyoucous hydrometer method according to Nelson and Sommers [28]. Samples were measured for both pH and electrical conductivity (EC) using a 1:2.5 (*v:v*) ratio of soil sample to deionized water with a 3020 pH meter (Jenway) and an EC meter (Jenway, 470 conductivity meter), respectively. Using the wet oxidation method, soil organic carbon (SOC) was determined according to [29]. Total nitrogen was determined in soil samples using the Kjeldahl method described by Olsen et al. [30]. Available phosphorus was determined according to the Olsen P method, based on the extraction of phosphate from the soil by sodium bicarbonate solution (0.5 N) adjusted to pH 8.5 [31]. The NH_4OAC extraction-flame photometry method was used to examine the availability of potassium in soil samples [32]. Physicochemical properties of soil before planting are shown in Table 1.

2.8. Statistical Analyses

All data were statistically analyzed according to the analysis of variance (ANOVA) and the least significant difference (L.S.D) method was used to compare differences among mean values according to the methods described by [28]. MSTAT-C Computer software Version 4 was used. The significant differences between treatment means were determined at $p \leq 0.05$ by using the Duncan test.

3. Results

3.1. Growth and Yield Characters

3.1.1. Potato

Growth of potato plants was good in all the studied characters with some variations according to the individual treatments. Some crop characteristics values increased with the applied treatments, and others decreased with the same treatments.

Plant production and yield per hectare

Data illustrated in Figure 2 show that there was a significant increase in plant production (yield/plant) in the T2 (767 g of fresh tubers) compared to the T4 treatment (control) (725 g), while the lowest plant production was in T3 (512.6 g), and production of potato was 623.6 in the T1 treatment. The total yield of fresh potato tubers/ha was 37.0 t ha⁻¹ in all plants treated with T2 and T4, with insignificant differences between the two values, while the lowest value resulted from plants treated with T3 (23.0 t ha⁻¹) (Figure 2).

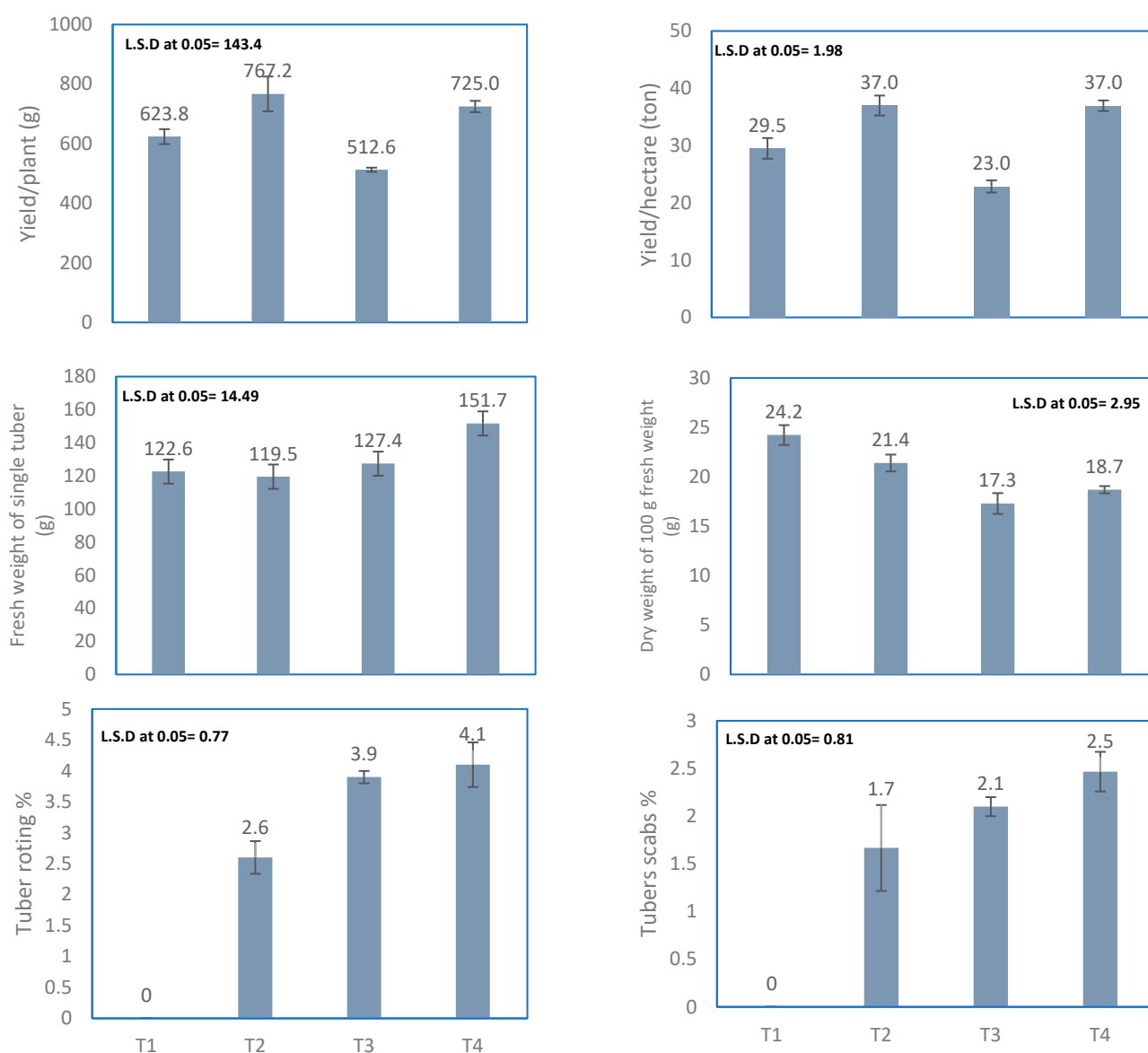


Figure 2. Effect of EMs, mycorrhizae biofertilizers as biostimulants and mineral fertilizers of NPK (75% and 100% control) on growth and productivity of potato under the Middle Egypt region conditions.

Fresh Weight of Single Potato Tubers and Dry Weight of 100 g Fresh Weight

Potato plants treated with the mineral fertilizers (NPK) (T4) gave the significant highest values of single tuber fresh weight (151.7 g) compared to plants treated with the biofertilizers, EMs and mycorrhiza (T1 and T2) (119.5 g and 122.6 g, respectively). Surprisingly, when 100 g of fresh potato slices was dried and weighed, the values were opposite, as the biofertilizers increased the dry matter content in the tubers. Potato plants fertilized with the biofertilizers (EMs and mycorrhiza) gave values of 21.4 and 24.2 g compared with the NPK treatment, which gave only 18.7 g of potato tuber dry weight (Figure 2).

Disease Infection in Produced Potato Tubers

Due to the treatment of potato plants with bio- and mineral fertilizers during the growing season, potato tuber rot and scab infection were analyzed in the produced tubers. Data in Figure 2 show that all obtained potato tubers from T1 (the mycorrhizae treatment) were clean and free from all rot and scab diseases (0.0% of rot or scab diseases), while those obtained from both (T2, T3 and T4) the EM and NPK treatments showed an infection (2.6%, 3.9% of tubers normal rot and 4.1%, respectively). For tuber scabs, the highest infection was in T4, with 2.5%, while the lowest infection was in T2, with 1.7%.

3.1.2. Onion

The applied bio- and mineral fertilizers significantly affected onion plant growth and total production per hectare along with affecting the bulb disease infection or bulb disorders and yield quality as described in the following recorded characteristics.

Plant Fresh Weight and Plant Length

The EM treatments (T2) significantly increased onion plants' fresh weight and gave an average value of 225.3 g compared to those of mycorrhizae and NPK treatments (T1, T3 and T4) which gave lower values (195.3, 185.4 and 208.8 g, respectively). However, using bio- or mineral fertilizers does not affect onion plant length as the average value was 44.3 cm in all treatments with insignificant differences among them (Figure 3).

Total Yield (t ha^{-1})

The total yield/hectare of onion was affected by the applied treatments. Both the 100% NPK and EM treatments (T2 and T4) have an insignificant increase on the total production of onion bulbs/hectare, giving mean values of 41.5 and 44.1 t ha^{-1} , respectively. The total yield of the mycorrhizae treatment (T1) was (36.8 t ha^{-1}), while the lowest yield was 29.5 t ha^{-1} in T3 as shown in Figure 3.

Dry Weight of 100 g of Onion Fresh Slices

As described in the potato section, the mycorrhizae treatment increased the dry matter content of onion bulbs insignificantly. The weight of 100 g of fresh onion slices obtained from this treatment was 12.2 g (average value), while those obtained from the EM and NPK treatments were 10.5 and 7.7 g, respectively. The NPK treatment insignificantly increased water content in onion bulbs of plants treated with these treatments (Figure 3).

Disorders in Onion Bulbs

Some disorders were noticed in bulbs produced from plants treated with bio- or mineral treatments, while plants treated with the mycorrhizae treatment showed no disorders.

Onion Bulb Rot and Sprouting

Onion plants fertilized with the NPK fertilizers (T3 and T4) gave bulbs infected with bulb rot (2.2% and 2.4%) and sprouting bulbs (5.5% and 6.3%), while those treated with the EM treatment gave only 1.2% with bulb rot and 3.4% sprouted bulbs. On the contrary,

plants treated with the mycorrhizae treatment gave onion bulbs with good quality and free from rot and only 2.7% sprouting bulbs (Figure 3).

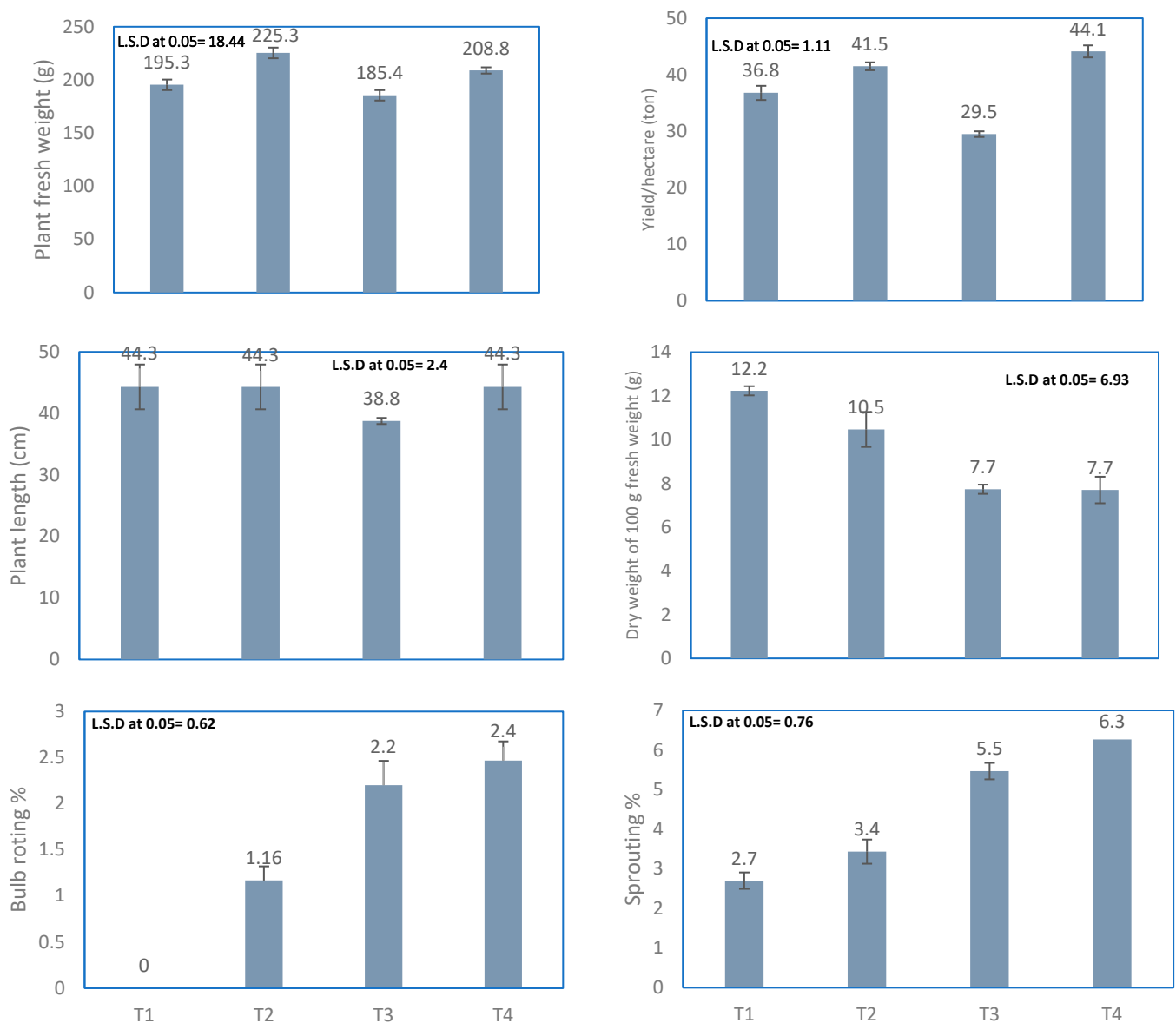


Figure 3. Effect of EMs, mycorrhiza biofertilizers and mineral fertilizers of NPK (75% and 100% control) on growth and productivity of onion under the Middle Egypt region conditions.

3.2. Effect of Mycorrhizae, Effective Microorganisms (EMs) and Doses Integrated with 75% of the Recommended NPK on Some Soil Properties

Data illustrated in Table 3 show the effect of effective microorganisms (EMs) or mycorrhizae integrated with 75% of the recommended NPK doses on some soil properties. In general, soil bulk density values varied from 1.15 to 1.23 Mg m⁻³, showing a tendency to decrease in all treatments as compared to control treatments with no significant difference except for soil treated with T2 (EMs + 75% NPK), where the bulk density was 1.15 Mg m⁻³. The soil pH was in the range between 8.08 and 8.16, which decreased significantly when affected by T1 (mycorrhizae applications). However, EMs had no significant effect on soil pH under the conditions of this experiment ($p < 0.05$). At the end of the experiment, the electrical conductivity (EC_e) was decreased in comparison to the soil before planting (Table 1). The obtained data showed that EC_e values varied from 3.31 to 3.98 dS m⁻¹. It was observed that the T4 (100% NPK) treatment recorded the highest EC_e value, 3.98 dS m⁻¹,

and the EC_e values were decreased significantly in the rest of treatments ($p < 0.05$). The most effective treatment in decreasing soil electrical conductivity was T2 (EMs + 75% NPK) with an EC_e value of 3.31 dS m^{-1} .

Table 3. Effect of the integration of mineral and biofertilization on soil properties planted with onion and potato crops.

Treatment	Dose	Bulk d. (Mg m^{-3})	pH 1:2.5 (Water)	EC_e (dS m^{-1})	SOC (g kg^{-1})	TN (g kg^{-1})	ava. P (g kg^{-1})	ava. K (g kg^{-1})
T1	Mycorrhizae + 75%NPK	$1.18 \pm 0.03 \text{ ab}$	$8.08 \pm 0.02 \text{ b}$	$3.66 \pm 0.14 \text{ b}$	$10.02 \pm 0.17 \text{ a}$	$1.01 \pm 0.07 \text{ b}^c$	$24.09 \pm 0.73 \text{ c}$	$275 \pm 9.42 \text{ a}$
T2	EM + 75% NPK	$1.15 \pm 0.04 \text{ b}$	$8.16 \pm 0.02 \text{ a}$	$3.31 \pm 0.13 \text{ c}$	$9.09 \pm 0.13 \text{ c}$	$0.97 \pm 0.07 \text{ c}$	$26.12 \pm 0.61 \text{ b}$	$226 \pm 4.58 \text{ b}$
T3	75% NPK	$1.22 \pm 0.04 \text{ a}$	$8.16 \pm 0.02 \text{ a}$	$3.51 \pm 0.10 \text{ b}^c$	$8.69 \pm 0.15 \text{ d}$	$1.12 \pm 0.04 \text{ ab}$	$21.00 \pm 0.42 \text{ d}$	$175 \pm 8.19 \text{ d}$
T4	100% NPK	$1.23 \pm 0.04 \text{ a}^*$	$8.13 \pm 0.02 \text{ a}$	$3.98 \pm 0.09 \text{ a}$	$8.71 \pm 0.23 \text{ d}$	$0.98 \pm 0.06 \text{ c}$	$23.98 \pm 0.68 \text{ c}$	$200 \pm 9.54 \text{ c}$

* Different letters indicate significant differences among means of treatments according to Duncan test at $p < 0.05$.

Soil organic carbon for various treatments ranged between 8.69 and 10.02 g kg^{-1} (Table 3). There was no significant difference between SOC content of T4 control (100% NPK) and T3 control (75% NPK) treatments. Application of EMs or mycorrhizae with 75% recommended doses of NPK fertilizers significantly increased SOC in comparison to the control treatments; particularly, mycorrhizae resulted in the highest SOC content, 10.02 g kg^{-1} . On the one hand, total nitrogen content for various treatments as presented in Table 3 showed no significant differences among T1, T2 and T4 (100% NPK, EMs + 75% NPK and mycorrhizae + 75% NPK treatments). Regarding available phosphorus, in comparison to the control (75% NPK), the combination of EMs or mycorrhizae with 75% NPK (T1, T2) was significantly higher in available phosphorus ($p < 0.05$). The highest available phosphorus value was observed in the T2 treatment, 26.12 g kg^{-1} . Integration between effective microorganisms (EMs) or mycorrhizae and 75% of the recommended NPK doses significantly increased the available potassium in comparison to the control (75% NPK) treatment ($p < 0.05$). The highest available potassium value was observed in T1 (mycorrhizae + 75% NPK), 275 g kg^{-1} .

3.3. Effect of Bio- and Mineral Fertilizers on Total Counts of Bacteria, Fungi, Actinomycetes and Mycorrhizal Colonization

Data presented in Table 4 show the effect of bio- and mineral fertilizers on the total counts of bacteria, fungi, actinomycetes and mycorrhizal colonization. The highest values of total counts of bacteria, fungi and root colonization were recorded with T1 (mycorrhiza + 75% NPK) followed by T2 (EMs + 75% NPK) in potato and onion as well. On the other hand, the highest values of total counts of actinomycetes were obtained with T2 (EMs + 75% NPK) followed by T1 (mycorrhiza + 75% NPK). The lowest values were obtained when using NPK alone without biofertilizers. For mycorrhizal colonization, data indicated that the percentage of mycorrhizal colonization after adding 75% NPK and mycorrhizal inoculation increased from 14% at zero fertilization (before planting) to 82% for potato plants and from 10% to 88% for onion plants. Colonization of roots is illustrated in Figures 4–6.

Table 4. Effect of bio- and mineral fertilizers on the total counts of bacteria, fungi, actinomycetes and mycorrhizal colonization in the soil samples planted with onion and potato.

Treatment	Potato				Onion			
	Bacteria ($\times 10^5$ CFU g^{-1})	Fungi ($\times 10^4$ CFU g^{-1})	Actinomycetes ($\times 10^5$ CFU g^{-1})	Mycorrhizal Colonization (%)	Bacteria ($\times 10^5$ CFU g^{-1})	Fungi ($\times 10^4$ CFU g^{-1})	Actinomycetes ($\times 10^5$ CFU g^{-1})	Mycorrhizal Colonization (%)
T1	188	60	81	82	200	66	87	88
T2	201	50	57	65	177	35	56	55
T3	144	45	95	32	152	54	88	40
T4	140	38	42	33	147	41	52	28
Before planting	88	15	12	14	78	14	14	10

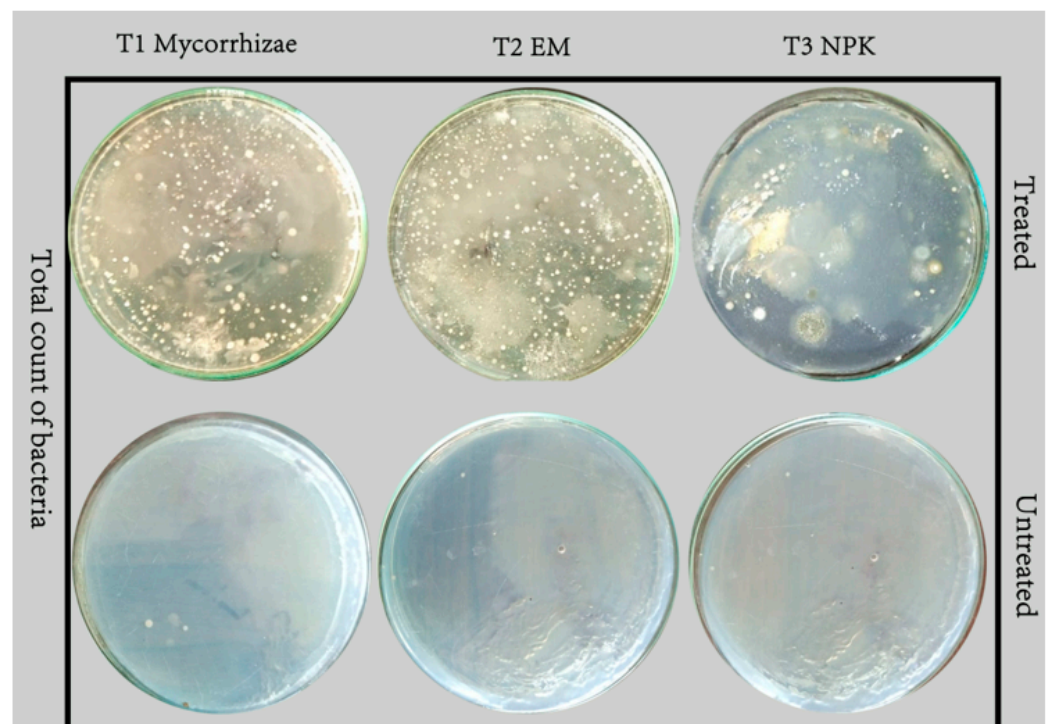


Figure 4. Effect of mycorrhizae, EMs and mineral fertilizers (NPK) on the total counts of bacteria in the soil planted with onion and potato (untreated dishes represent 100% NPK).

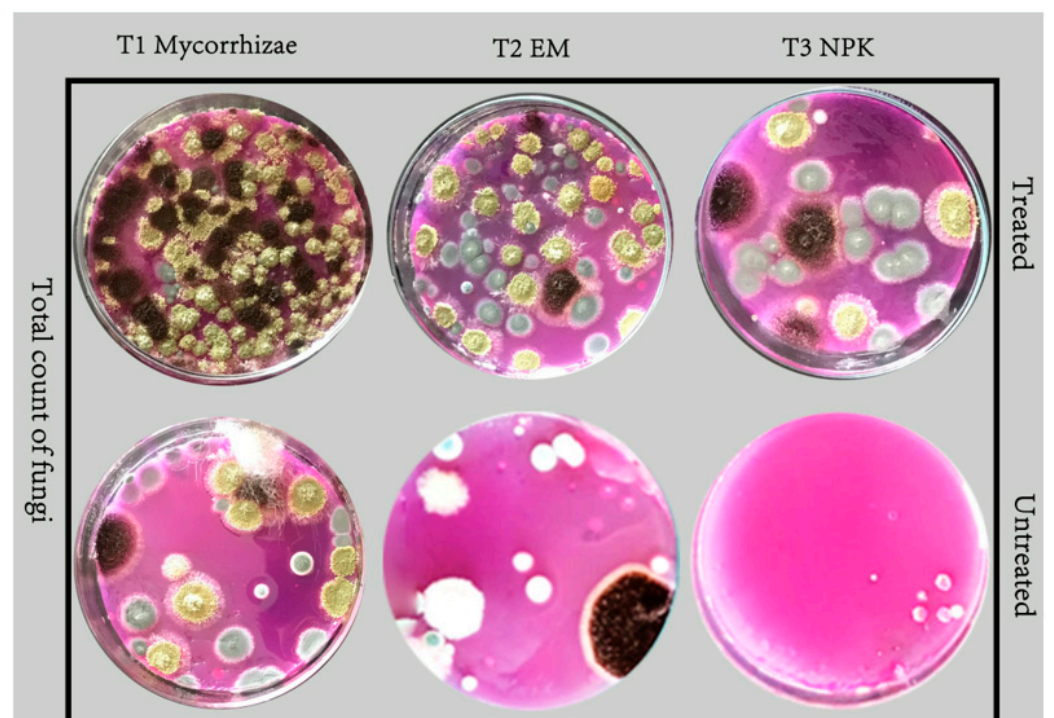


Figure 5. Effect of mycorrhizae, EMs and mineral fertilizers (NPK) on the total counts of fungi in the soil planted with onion and potato.

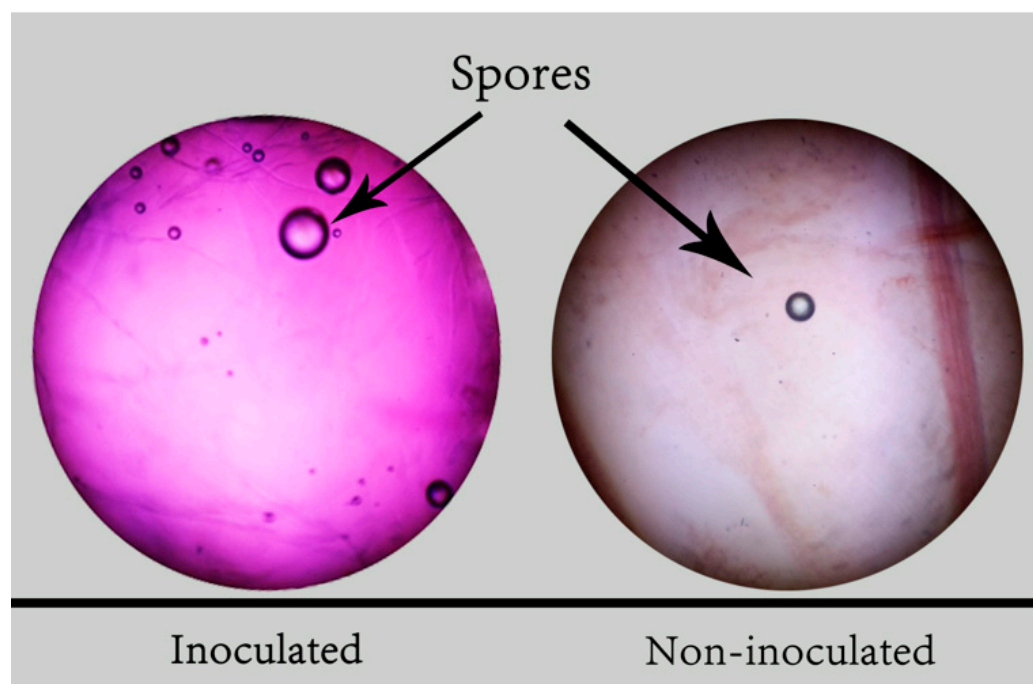


Figure 6. Mycorrhizal colonization (root sections for inoculated and non-inoculated samples).

4. Discussion

With the huge increase in global population and demand for food, chemical fertilizers are gradually beginning to play an important role in crop productivity maintenance, in addition to herbicides and pesticides, which are significantly important for maintaining crop health. Currently, public concern has been focused on food security issues and environmental pollution problems which are caused by the excessive application of herbicides, fertilizers and pesticides. Biofertilizers have recently been extensively studied and applied as possible substitutes for chemical fertilizers because of their strong progression of plant growth, soil improvement, disease control, etc. [33].

In Egypt, the use of biofertilizers is limited by the threshold of permissible nitrogen level (170 kg N ha^{-1} per year) according to EU regulation numbers 834/2007 and 889/2008. The limitations are especially significant for the export of organic products; thus, biofertilizer use was focused on through our study with the aim of decreasing the rate of fertilizer. In this study, some plant biostimulating organisms were used, such as EMs and mycorrhiza in plant protection, as a possible means of ensuring present and future food security. Biostimulants could be a promising tool in the current scenario of crop production. They are beneficial microorganisms of natural sources that can promote plant growth under various environmental stresses [34–37]. This was proved in our experiments as these microorganisms could promote onion and potato growth, yield and quality of the obtained production of onion bulbs and potato tubers. They also decreased or eliminated the disorders in these bulbs and tubers.

Biofertilizers contain latent or live microorganisms that can mobilize nutrients from unavailable to available form via many biological processes [38]. Proper use of biofertilizers helps in preserving the fertility of agricultural soils [39]. Numerous studies reported the positive impacts of biofertilizer on plant growth and crop yield compared to the control [40–44]. Our results showed that potato yield was increased by inoculation with bacteria in comparison to the control treatment. The reliable explanation for this is that the beneficial microorganisms found in the biofertilizers such as *Bacillus* and *Azotobacter* can promote plant growth. *Azotobacter* spp. also can assist plant resistance to many pathogens that may infect plants by producing antifungal compounds which cause promotion of plant productivity [45]. *Azotobacter* and *Azospirillum* can colonize near and around the root zone,

and they can promote nitrogen availability in the soil through N fixation. Nitrogen is an essential macronutrient to plants as it ameliorates the growth of the shoot system, supports reproduction and represents the main constituent of chlorophyll, which is responsible for the green color and photosynthesis [46]. The enhanced potato yield as a result of using biofertilizers might be due to the promotion of root growth and functions, enhanced uptake of minerals into the plant and production of phytohormones such as cytokinin and IAA GAs, which also lead to the reduction in abscisic acid [47]. Our results are in harmony with those of Zaghoul [48], who reported that the growth parameters were significantly increased in the inoculated potato tuber treated with *Azotobacter* spp. and *Azospirillum* spp. through the production of some phytohormones such as cytokinin, auxin, abscisic acid and gibberellins, which can enhance plant productivity and promote plant growth [49].

For onion plants, the obtained results of this investigation indicated that yield and bulb weight were significantly increased by biofertilizers and biominerals. The role of biofertilizer application in improving growth and yield of onion may be due to the increased availability of nitrogen for absorption to onion plants, which increase the nitrate accumulation on onion bulbs [50–52]. These impacts could be attributed to the increases in root hairs, root elongation and root surface area as mentioned by Sundaravelu and Muthukrishnan [53]. These results are in harmony with the results of Yogita and Ram [54] on onion. The yield was higher in EMs than mycorrhiza, while contrasting results were found for the dry matter. The results showed that biofertilizers have a better impact on plant length compared to mineral fertilizers. This may be attributed to *Azospirillum*, *Azotobacter* and *Bacillus*, which, in addition to being able to fix nitrogen, can produce exopolysaccharides and phytohormones such as auxins and gibberellins, which cause an increase in values of onion height. Our gained results are in good agreement with Yadav et al. [55] on onion and Pradu et al. [56] on okra.

Fresh weight of potato and onion showed no significant differences in all the treatments compared to the control, while dry weight increased significantly in both EMs and mycorrhiza with 75% NPK. The recorded data for both fresh and dry weight showed the useful impact of biofertilizers and their influence in creating preferable soil conditions for chemical, physical and microflora properties of the soil. In addition to the ability of biofertilizers to enhance soil status, they also eliminate the environmental pollution and hazards associated with using chemical fertilizers. Our results agreed with Oyeyiola [57] and Himanni et al. [58].

In potato production, a major limiting factor is disease, which can be soilborne, seed-borne or both. Among soil microorganisms, fungi and bacteria have received great attention as biocontrol agents for mitigating soil borne pathogens that infect plants [59,60]. Lately, biological control agents have been investigated for plant diseases as alternatives to synthetic pesticides because of their lower environmental impact, enhanced degree of safety and ability to reduce disease [61]. Rot, caused by the *E. carotovora* bacteria subsp. *carotovora*, is considered the most dangerous of all storage diseases. Our results suggest that the use of biofertilizers can reduce rot disease, which appeared significantly in the control treatments. Our results also detected that the highest fresh weight for potato was obtained due to the control treatment. The highest dry weight was obtained due to the treatment with mycorrhiza, while EM treatment gave the lowest fresh weight. These findings are similar to the results reported by Couillerot et al. [62], who pointed out that scab disease does not affect or reduce yields, but it may degrade the quality and appearance of tubers, which is particularly important when growing potatoes for nourishment. Biofertilizers and biotreatments may be a beneficial tool for the management of disease in Egypt.

During storage of onion in the field, it can be infected by some fungal and bacterial diseases, which can cause severe loss. The most destructive disease is black rot, which is caused by *Aspergillus niger* [63]. The data showed that all the treatments increased the tolerance of onion against rot and sprouting diseases and maintained the bulbs healthy bulbs during storage process.

The microbial population that is present in the rhizosphere is different from that surrounding it due to the root exudates that act as a nutrition source for growth of microbes [64]. Microbes colonize the interior of the plant or the rhizosphere and improve growth by increasing primary nutrient availability of to the host plant through phosphorus solubilization, biological nitrogen fixation and stimulating plant growth via the synthesis of substances responsible for growth promotion. Our results reflected that the addition of bio- and mineral fertilizers improves the microorganisms in the soil and the microbial count. The results correlate with those of Mandic et al. [65] and Javoreková et al. [66], who reported that adding these substances resulted in microbial activity enhancement.

Effective microorganisms can improve soil quality [67,68]. Gang et al. [69] mentioned that soil treated with microorganisms can improve soil structure and make it less compact, better drained and more friable. This could lead to improved growing conditions for plant growth. Our results showed an increase in the soil organic carbon (SOC) in the soil treated by EMs and mycorrhiza. Soil organic carbon is directly related to microbial population of the soil [70]. The increase in SOC will also result in increased nutrient availability for the growing plant by augmenting the biological activity of the soil, which causes enhanced nutrient use efficiency. For potassium and phosphate content, soil treated with biofertilizers had higher P and K content than mineral fertilizers. These results proved the presence of a direct relationship between inoculation content and the available nutrients of the soil. Addition of biofertilizers could increase the P availability in soil, as it can sustain a greater population of bacteria which are able to solubilize soil P. Application of biofertilizers leads to solubilization of insoluble P in soil and thus higher uptake of P [71]. Our results are in harmony with those of Ramalkashi [72]. According to Hamdia et al. [73], biofertilizers with noticeably changed soil ion selectivity enhance K^+ absorption. It was claimed that the use of biofertilizers resulted in the buildup of N, P and K individually, maintaining the nutritional balance [74,75].

Present findings showed that soil pH values were reduced with biofertilizer application. The shift in soil pH may be due to the increase in organic acid levels with microbial inoculation, which reduces soil pH. Present findings agree with the findings of Gopinath et al. [76] and Jaipaul et al. [77]. Berger et al. [78] reported that biofertilizers decreased soil pH and raised available P and K levels in the soil mainly due to increased release of K from minerals and organic compounds.

For bulk density of the soil, the results showed a decrease in bulk density of soil treated with biofertilizers compared to mineral treatments, which may be due to trapping of organic fractions within soil aggregates. The decreases in soil bulk density with biofertilizer treatments were primarily attributed to the reduction in specific gravity of organic materials, improved aggregation and raised soil volume [79]. Present results on bulk density agreed with those of Abd El-Ghany et al. [80] and Li et al. [81] reporting decreased soil bulk density with biofertilizer treatments. Soil electrical conductivity also decreased with biofertilizer application. This may be due to the positive effect of microorganisms on soil organic matter, which affects soil structure and water movement in soil and accordingly salt leaching from the top surface soil layer. Our findings were in harmony with those of Zeynep [82] and Hala et al. [83].

5. Conclusions

Field and laboratory (soil and microbiology) experiments were conducted to examine the positive and negative effects of microorganisms (EMs and mycorrhiza) and NPK fertilizers on growth, production and yield quality of onion and potato products as well as to show the relationship of those treatments with the environment and climate change in the Middle Egypt region. Results showed that the used biofertilizer microorganisms increased onion and potato plant growth and dry matter yield. They also increased the quality of the potato tubers and onion bulbs and decreased the disorders of these products when compared with the mineral treatment (NPK fertilizers). Moreover, the biofertilizers (EMs and mycorrhizae) increased the content of different beneficial microorganisms in the

soil and enhanced the soil properties as described above. Furthermore, those biofertilizers reduced the environmental stresses affected onion and potato plant growth and production and had a part in protecting the environment where the onion and potato plants were grown from the negative effects of climate change. No chemical pesticides for soilborne or airborne diseases were used in this study as they are very harmful for plants and the environment. The biofertilizers used in the present study reduced the climate change stress on these vegetable crops in the Middle Egypt region and could do the same in similar locations. Moreover, using these biocontrol agents decreased the total cost of producing onion and potato as they decreased the cost of fertilization and pesticides by 25%.

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